Status of the Claims:

1. (currently amended) A composition for determining the presence or absence of a target molecule comprising a first ribonucleic acid (RNA) molecule, said first RNA molecule binds a target molecule and has the following formula:

5'-A-B-C-D-E-3';

wherein A is a section of the RNA molecule having 10-100,000 nucleotides which section is, with another RNA sequence, E, replicated by an RNA replicase, the letter "B" denotes a section of the RNA molecule having approximately 1 to 50000 nucleotides which section, with another sequence D, binds the target molecule under binding conditions, wherein said target is a small or large organic molecule ~~such as~~ selected from the group consisting of a peptide, protein, and derivatives thereof, the letter "C" denotes a section of the RNA molecule having approximately 1 to 10000 nucleotides which section is capable preventing the replication of the first molecule by the RNA replicase, the letter "D" denotes a section of the RNA molecule having approximately 1 to 50000 nucleotides which section, with another sequence B, binds the target molecule under binding conditions, the sections B and D, in combination, comprise in total at least 10 nucleotides, the first RNA molecule, with sections B and D bound to target, is acted upon by the RNA replicase to form a second RNA molecule, said second RNA molecule has the following formula:

5'-E'-X-A'-3';

wherein, E' is the complement to E, and A' is the complement to A, and the letter "X" denotes the complement of parts of the sections B and D which may be replicated, or the letter denotes the direct bond between sections E' and A', and said second RNA molecule is replicated by the RNA replicase under replicating conditions.

2. (original) The composition of claim 1 wherein the sections represented by the letters "A" and "E" are selected from the group of sequences consisting of MDV-I RNA, Q-beta RNA microvariant RNA, nanovariant RNA, midivariant RNA, RQ-135 and modifications of such sequences which maintain the ability of the sequences to be replicated by Q-beta replicase.

3. (original) The composition of claim 1 wherein the RNA replicase is Q-beta replicase.

4. (original) The composition of claim 1 wherein the sections B and D bind to target through non-nucleic acid base pairing interactions.

5. (original) The composition of claim 1 wherein the sections B and D each have a hybridization sequence of 1-100 nucleotides, said hybridization sequence of section B is

adjacent to the section A and forms a hybridization product with a said hybridization sequence of section D, and said hybridization sequence of section D is adjacent section E.

6. (original) The composition of claim 1 wherein the section C has 1-10,000 nucleotides which sequences define a stop sequence for the RNA replicase.

7. (original) The composition of claim 1 wherein the sections A and E comprise at least one sequence that hybridizes to a third nucleic acid to form a hybridization product which hybridization product can be detected.

8. (currently amended) A composition for determining the presence or absence of a target molecule comprising paired RNA molecules having a first RNA molecule and a second RNA molecule, said first RNA molecule binds a target molecule and has the following formula:

5'-A-F-B-3';

and, said second RNA binds the target and has the following formula:

5'-D-H-E-3';

wherein A is a section of the RNA molecule having 10-100,000 nucleotides which section is, with another RNA sequence, E, replicated by an RNA replicase, the letter "B" denotes a section of the RNA molecule having approximately 1 to 50000 nucleotides which section, with another sequence D, binds the target molecule under binding conditions, wherein said target is a small or large organic molecule ~~such as~~ selected from the group consisting of a peptide, protein, and derivatives thereof, the letter "D" denotes a section of the RNA molecule having approximately 1 to 50000 nucleotides which section, with another sequence B, binds the target molecule under binding conditions, wherein said target is a small or large organic molecule ~~such as~~ selected from the group consisting of a peptide, protein, and derivatives thereof, the sections B and D, in combination, comprise in total at least 10 nucleotides, the letter "F" denotes a section of the RNA molecule having has a hybridization sequence of 1-10,000 nucleotides which form a hybridization product with a section H , the letter "H" denotes a section of the RNA molecule having has a hybridization sequence of 1-10,000 nucleotides which form a hybridization product with a section F, in the absence of target, the hybridization sequences do not form a stable hybridization product, in the presence of the target, and the formation of a complex between sections B and D with the target, a hybridization product is formed that allows the RNA replicase to replicate sections A and E to form a third RNA molecule, said third RNA molecule has the following formula:

5'-E'-X-A'-3';

wherein E' is the complement to E and A' is the complement to A, the letter "X" denotes the complement of parts of the sections F and H which may be replicated, or the letter

denotes the direct bond between sections E' and A' and said third RNA molecule is replicated by the RNA replicase under replicating conditions.

9. (withdrawn)

10. (canceled)

11. (currently amended) A kit for determining the presence or absence of a target molecule, said kit comprises a one or more reagents comprising a first RNA molecule for use with an RNA replicase, said first RNA molecule has the formula:

5'-A-B-C-D-E-3';

wherein A is a section of the RNA molecule having 10-100,000 nucleotides which section is, with another RNA sequence, E, replicated by an RNA replicase, the letter "B" denotes a section of the RNA molecule having approximately 1 to 50000 nucleotides which section, with another sequence D, binds the target molecule under binding conditions, wherein said target is a small or large organic molecule ~~such as~~ selected from the group consisting of a peptide, protein, and derivatives thereof, the letter "C" denotes a section of the RNA molecule having approximately 1 to 10000 nucleotides which section is capable preventing the replication of the first molecule by the RNA replicase, the letter "D" denotes a section of the RNA molecule having approximately 1 to 50000 nucleotides which section, with another sequence B, binds the target molecule under binding conditions, the sections B and D, in combination, comprise in total at least 10 nucleotides, the first RNA molecule, with sections B and D bound to target, is acted upon by the RNA replicase to form a second RNA molecule, said second RNA molecule has the following formula:

5'-E'-X-A'-3';

wherein, E' is the complement to E, and A' is the complement to A, and the letter "X" denotes the complement of parts of the sections B and D which may be replicated, or the letter denotes the direct bond between sections E' and A', and said second RNA molecule is replicated by the RNA replicase under replicating conditions, said kit for determining the presence or absence of said target molecule.

12. (currently amended) A kit for determining the presence or absence of a target molecule comprising paired RNA molecules said paired RNA molecules comprising a first RNA molecule and a second RNA molecule, said first RNA molecule has the formula:

5'-A-F-B- 3'.

The second RNA molecule has the formula:

5'-D-H-E-3'

wherein A is a section of the RNA molecule having 10-100,000 nucleotides which section is, with another RNA sequence, E, replicated by an RNA replicase, the letter "B" denotes a section of the RNA molecule having approximately 1 to 50000 nucleotides which section, with another sequence D, binds the target molecule under binding conditions, wherein said target is a small or large organic molecule ~~such as~~ selected from the group consisting of a peptide, protein, and derivatives thereof, the letter "D" denotes a section of the RNA molecule having approximately 1 to 50000 nucleotides which section, with another sequence B, binds the target molecule under binding conditions, wherein said target is a small or large organic molecule ~~such as~~ selected from the group consisting of a peptide, protein, and derivatives thereof, the sections B and D, in combination, comprise in total at least 10 nucleotides, the letter "F" denotes a section of the RNA molecule having has a hybridization sequence of 1-10,000 nucleotides which form a hybridization product with a section H , the letter "H" denotes a section of the RNA molecule having has a hybridization sequence of 1-10,000 nucleotides which form a hybridization product with a section F, in the absence of target, the hybridization sequences do not form a stable hybridization product, in the presence of the target, and the formation of a complex between sections B and D with the target, a hybridization product is formed that allows the RNA replicase to replicate sections A and E to form a third RNA molecule, said third RNA molecule has the following formula:

5'-E'-X-A'-3';

wherein E' is the complement to E and A' is the complement to A, the letter "X" denotes the complement of parts of the sections F and H which may be replicated, or the letter denotes the direct bond between sections E' and A' and said third RNA molecule is replicated by the RNA replicase under replicating conditions.

13. (withdrawn)

Remarks

It may be helpful to outline the essentials of the presently claimed invention before addressing the Examiner's individually stated rejections. The instant invention relates to the detection of target proteins in a sample. A first RNA, putatively specific for the protein target, interacts and binds to the target protein. When the first RNA binds to the target protein, the *cis* regulatory element of the first RNA (the "C" sequence) is rendered inoperable thereby allowing for the first RNA to serve as a template for RNA synthesis. A second RNA is synthesized, using the first RNA as a template, such that it is complementary to the first RNA. However, should the first RNA NOT interact with and bind to a target protein, then the "C" *cis* regulatory element will prevent synthesis and thus the formation of the second RNA molecule.

Claims 1-8, 11 and 12 are rejected under 35 USC § 112, second paragraph

Claims 1-8, 11 and 12 are rejected under 35 USC § 112, second paragraph, as being indefinite for failing to particularly point out and distinctly claim the subject matter which Applicants regard as the invention.

The amendments presented above should alleviate the concern of the Examiner. The Examiner specifically alludes to the use of the phrase "such as" in the claims. This phrase has been deleted in the amended claims. Applicants respectfully request reconsideration and withdrawal of the present rejection.

Claims 1-8 are rejected under 35 USC § 103(a)

Claims 1-8 are rejected under 35 USC § 103(a) over Marsh *et al.* in view of Spiegelmen further in view of Holy *et al.* (US Pat. No. 5,977,061). Applicants respectfully disagree.

The Examiner posits that Marsh *et al.* teach a method of determining the presence or absence of a target molecule. Further, that Marsh *et al.* [*sic*] "does not teach a

composition by providing paired RNA molecules. Marsh *et al.* does not teach section "C" of the RNA molecule which section is capable of preventing the replication of the first molecule by the RNA replicase." *Office Action* dated 8/14/03, pg. 5.

The Examiner further characterizes Spiegelman as teaching [*sic*] "a customized preparation of RNA templates ... Spiegelman teaches section "C" of the RNA molecule which section is capable preventing the replication of the first molecule by the RNA replicase." *Office Action* dated 8/14/03, pp. 5-6.

The Examiner states that [*sic*] "Holy *et al.* teaches the nucleic acid, wherein the target is a small or large organic molecule such as a peptide, protein, and derivatives thereof, which can be attached to the analogues of known bases or nucleotides." *Office Action* dated 8/14/03, pg. 7.

In order to establish a *prima facie* case of obviousness, "there must be some suggestion or motivation, either in the references themselves or in the knowledge generally available to one of ordinary skill in the art, to modify the reference or to combine reference teachings. Second, there must be a reasonable expectation of success. Finally, the prior art reference (or references) must teach or suggest all of the claim limitations." M.P.E.P. §2143, see also, *In re* Vaeck, 947 F.2d 488, 20 USPQ2d 1438 (Fed. Cir. 1991).

A case of *prima facie* obviousness is not been established. The presently claimed invention claims a composition for determining the presence or absence of a target molecule comprising a first RNA molecule that binds the target molecule of interest. The first RNA molecule has the formula of:

5'-A-B-C-D-E-3'

wherein A is a section of the RNA molecule together with section E is replicated by a replicase; "B" denotes a section of RNA sequence together with "D" that actually binds the target molecule under proper binding conditions; "C" is a section of RNA that is capable of preventing replication. Once the target molecule is bound by sections "B" and

"D" a replicase acts upon the complex to form a second RNA molecule having the following formula:

5'-E'-X-A'-3'

wherein, E' is the complement to E, and A' is the complement to A, and "X" denotes the complement of parts of "B" and "D" that can be replicated, or the direct bond between sections E' and A'.

Applicants claim a composition comprising paired RNA molecules having a first RNA molecule and a second RNA molecule. The first RNA molecule binds a target molecule and has the following formula:

5'-A-F-B-3'

and a second RNA molecule that binds a target molecule having the following formula:

5'D-H-E-3'

wherein, A and E are subject to replication by a replicase, "B" and "D" denote sections responsible for binding to a target molecule, "F" forms a hybridization product with section "H" that allows for replication of sections A and E to form a third RNA molecule having the formula:

5'-E'-X-A'-3'

wherein E' is the complement to E and A' is the complement to A, "X" denotes the complement of parts of "F" and "H" that can be replicated, alternatively, "X" denotes the direct bond between E' and A' and this third RNA molecule is subject to replication.

As stated above, a requirement to establish a *prima facie* case of obviousness requires that the prior art reference (or references) must teach or suggest all of the claim limitations - this requirement is simply not met by Marsh either alone or in combination with Spiegelman and/or Holy.

As stated above, Marsh fails to recite an RNA molecule having the formula 5'-A-B-C-D-E-3' nor does Marsh disclose an RNA having the formula 5'-A-F-B-3' nor 5'-D-H-

E-3' with the attendant limitations of these formulae. For example, there is no analog to RNA sections "B" and "D" as claimed by Applicants in the Marsh disclosure. Recall, these sections in Applicants' invention form secondary structure and are employed to bind a target molecule, specifically a protein target. Marsh fails to recite any analogous RNA regions. Still another example, "F" and "H" which serve to form a hybridization product in Applicants' claimed invention is completely absent in Marsh. Further, unlike the presently claimed invention, Marsh fails to disclose a paired RNA molecule composition.

Moreover, section "C" as claimed by Applicants is completely absent from Marsh, as adroitly pointed out by the Examiner on page 5 of the *Office Action* dated 8/14/03. In the presently claimed invention, section "C" is a stretch of RNA that can be used to inhibit replicase activity. The Examiner points out that Marsh has an analogous section that serves as a non-base-paired spacer to facilitate access of the replicase to the promoter. This simply is not equivalent to that which is presently claimed. Applicants in the specification characterizes section "C" as comprising "stop" sequences. This is very different than just a spacer element. Therefore, Marsh fails to recite an equivalent to sequence "C" of Applicants' presently claimed invention.

Further, in order to arrive at the claimed second RNA molecule, a target (protein) molecule has to bind with sections "B" and "D" of the first molecule, then replicase activity takes place. There is no analogous disclosure to be found in Marsh *et al.*

Spiegelman fails to rectify the deficiencies of Marsh. Spiegelman fails to recite an RNA molecule having the formula 5'-A-B-C-D-E-3' nor does Speigelman disclose an RNA having the formula 5'-A-F-B-3' nor 5'-D-H-E-3' with the attendant limitations of these formulae. For example, there is no analog to RNA sections "B" and "D" as claimed by Applicants in the Spiegelman disclosure. Recall, these sections in Applicants' invention form secondary structure and are employed to bind a target molecule, specifically a protein target. Spiegelman fails to recite any analogous RNA regions. Still another example, "F" and "H" which serve to form a hybridization product in Applicants'

claimed invention is completely absent in Spiegelman. Further, unlike the presently claimed invention, Spiegelman fails to disclose a composition by providing a paired RNA molecule.

Moreover, section "C" as claimed by Applicants is completely absent from Spiegelman. In the presently claimed invention, section "C" is a stretch of RNA that is contiguous with the other sections of the RNA molecule and can be used to inhibit replicase activity. The Examiner suggests that Spiegelman has an analogous section. The "interfering compound" as Spiegelman puts it is an RNA molecule independent from the viral RNA molecule of interest. The interfering compound comprises nucleotide sequences that will interact with a replicase thus precluding replication of the independent viral RNA. This simply is not equivalent to Applicants' claimed invention. Section "C" in the presently claimed invention comprises "stop" sequences that is contiguous with the larger RNA molecule.

Further, in order to arrive at the claimed second RNA molecule, a target (protein) molecule has to bind with sections "B" and "D" of the first molecule, then replicase activity takes place. There is no analogous disclosure to be found in Spiegelman.

Holy *et al.* disclose nucleotides that can be used as intermediates in the formation of flame retardants, diagnostic reagents and therapeutics, including antivirals. See Abstract of '061. One embodiment of the '061 patent involves producing antibodies against a nucleotide of the invention. These antibodies can be labeled and be used to bind with analogues of the invention, thereby detecting it. See, '061, columns 12-13, lines 64 -2.

Holy is significantly different from the presently claimed invention. The target class of molecules in Holy are nucleotides as well as other proteins and the ligand is a protein, the produced antibodies. Whereas, in the presently claimed invention, the target molecules are proteins and the ligand is an RNA molecule.

Moreover, the detection mechanisms are quite different. In Holy, the antibody produced against a particular nucleotide is labeled and if, and when, it interacts with its target the complex can be detected. In the presently claimed invention a first RNA molecule must first bind to a target molecule, then and only then, is said first RNA molecule replicated forming a second RNA molecule. This replication of the first RNA forming the second RNA can serve as a signal for detecting the presence of the target in a given sample. The second RNA molecule can go on to be further replicated. A reiterative cycle can be envisaged, the reagents and replicase activity are obviously limiting.

The scenario disclosed in Holy is quite different what is claimed in the present invention. To reiterate, a first RNA molecule is used to interact with a target molecule, a protein, in a complex sample. If the target protein is present, then the first RNA will interact and bind thereto. Once bound, the first RNA molecule can serve as a template for the synthesis of a second RNA molecule. The production of the second RNA molecule can then serve as a signal amplification event. If, however, the target protein is absent from the sample, then, the first RNA will NOT serve as a template for RNA synthesis. In Holy, the antibody serves as the ligand, whereas in the presently claimed invention it is the first RNA molecule that serves as the ligand. (Here ligand is understood to mean that molecule that interacts with a target molecule.) Clearly, Holy is significantly different from the presently claimed invention. Applicants respectfully request reconsideration and withdrawal of the present reject.

Claims 11 and 12 are rejected under 35 USC § 103(a)

Claims 11 and 12 are rejected under 35 USC § 103(a) over Marsh *et al.* in view of Spiegelmen further in view of Holy *et al.* (US Pat. No. 5,977,061) further in view of the Stratagene Catalog. Applicants respectfully disagree.

The deficiencies of Marsh and Spiegelmen have been presented above and need not be repeated here. The arguments presented above apply equally here as well.

As stated previously, Holy *et al.* disclose nucleotides that can be used as intermediates in the formation of flame retardants, diagnostic reagents and therapeutics, including antivirals. See Abstract of '061. One embodiment of the '061 patent involves producing antibodies against a nucleotide of the invention. These antibodies can be labeled and be used to bind with analogues of the invention, thereby detecting it. See, '061, columns 12-13, lines 64 -2.

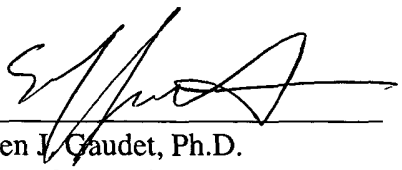
As described above, Holy is significantly different from the presently claimed invention. The target class of molecules in Holy are proteins as well as nucleotides and the ligand is a protein, the produced antibodies. In the present invention, the target class of molecules include protein molecules and the ligand is a nucleotide molecule. (See previous discussion for detail arguments against Holy.)

The Stratagene Catalog fails to rectify the deficiencies found in the cited references. The Stratagene Catalog does disclose various kits having various reagents, however, this is wholly insufficient to establish a case of *prima facie* obviousness. In order for Stratagene to be effective in establishing a case of obviousness, it would have to rectify the deficiencies found in the other cited references. Moreover, there would have to be a suggestion or motivation for one to combine Stratagene with any of the other cited references. This suggestion or motivation is completely lacking in either reference.

Marsh *et al.* either alone or in combination with Spiegelman and/or Holy does not establish a *prima facie* case of obviousness. Moreover, there is a paucity of motivation to combine these references and even if one did, there is no reasonable expectation of success that by such combination one would arrive at the presently claimed invention. Therefore, Applicants respectfully request reconsideration and withdrawal of this rejection.

The Examiner is invited to call the undersigned attorney at (617) 854-4237 should he determine that a telephonic interview would expedite prosecution of this case.

Respectfully submitted,



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Date: 9/2/03